

Lack of polymyxin resistance among carbapenemase-producing *Enterobacteriaceae* in a university hospital in China

Sir,

We read with interest two recent articles in the present journal focusing on escalation of carbapenemase-producing *Enterobacteriaceae* in the hospital environment, one from the Zhejiang Province, China [1], and another from Northern Italy [2]. Multidrug resistance is increasingly reported in clinical enterobacterial isolates in China. Not only resistance to carbapenems is on the rise, but in addition co-resistance to other clinically-relevant antibiotics such as fluoroquinolones, aminoglycosides and fosfomycin is increasingly observed. Polymyxins are therefore considered as last resort antibiotics for treating infections due to multidrug-resistant Gram negatives.

Recently, plasmid-mediated colistin resistance (MCR-1) was reported in enterobacterial from animals, food and patients from China [3]. Beyond China, the spread of the *mcr-1* gene has been now reported worldwide in many enterobacterial species [4]. In addition, one of the major concern is the spread of carbapenem- and polymyxin-resistant nosocomial *Klebsiella pneumoniae* isolates as observed in Italy [5] and more recently in France [6]. High plasmid-mediated *mcr-1* carriage rate has been observed in *E. coli* collected from raw meat (15%) and animals (21%) in China, but not from inpatients (1%) [3]. Recently, a variant of MCR-1, namely MCR-2, has been reported from Belgium [7].

Taking into account the wide spread MCR-1-producing strains in China and elsewhere, the diversity of genetic structures associated to the *mcr-1* gene identified so far, and the diversity of the clonal backgrounds of the strains harboring that gene, it is likely that its spread does not correspond to a recent event in China.

We have performed a retrospective study to evaluate the occurrence of polymyxin resistance and of the

MCR-1 determinant among carbapenemase-producing *Enterobacteriaceae* isolates, including 112 *K. pneumoniae*, 15 *Escherichia coli*, 15 *Citrobacter freundii*, 12 *Enterobacter aerogenes*, 3 *Klebsiella oxytoca*, 2 *Citrobacter farmeri*, a single *Enterobacter cloacae* and a single *Citrobacter braakii*. Those clinical isolates have been recovered during the 2006–2011 period at the Huashan Hospital of Fudan University, Shanghai. Ninety-four were from sputum, 43 from urine, 6 from drainage, 5 from blood, 3 from cerebrospinal fluid and 10 from other samples. Carbapenemase activity was assessed using the Rapidec Carba NP test (bioMérieux, La Balme-les-Grottes, France) [8]. Carbapenemase genes were searched as previously described [7]. All isolates were screened for polymyxin resistance by using the recently-developed Rapid Polymyxin NP test [9]. MICs of colistin and polymyxin B were determined by broth microdilution method as recommended by CLSI [10]. All polymyxin-resistant isolates were screened for the *mcr-1* and *mcr-2* genes by using PCR with primers *mcr-all-F* (5'-TATCGCTATGTGCTAAAG-3') and *mcr-all-R* (5'-TCTTGGTATTTGGCGGTA-3'). All of 161 isolates were tested positive for the Rapidec Carba NP test. Further PCR and sequencing showed that all strains were positive for *bla*_{KPC-2}. No polymyxin-resistant isolate was detected using the Rapid Polymyxin NP test, and further determination of MICs of colistin and polymyxin B confirmed that all isolates were susceptible to polymyxins. Accordingly, all isolates tested negative for the *mcr-1* and *mcr-2* genes.

This study revealed the absence of polymyxin-resistant isolate in our collection of carbapenemase-producing enterobacterial isolates. Combining with results obtained through other Chinese studies [4,6,8], our data suggest that polymyxin resistance rates may be much lower than expected in Chinese hospitals. However, surveillance of

polymyxin resistance shall be implemented on a regular basis to monitor its potential emergence in China.

To conclude, polymyxins might still constitute clinically-relevant options for treating infections caused by carbapenemase-producing Enterobacteriaceae in China. However, the use of polymyxins for humans is still not approved in China, considering its significant toxicity.

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Disclosure statement

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